Immunoperoxidase Staining

An antibody is an immunoglobulin molecule produced by the immune system that has the property of being able to combine with specific molecules called antigens. This binding is highly specific for the species and is dependent upon the selection process involved in soliciting antibody production. Antigens can be found intra and extracellular and in many cases convey a uniqueness to a cell or a particular group of cells. It is this uniqueness that allows pathologists to identify certain diseases or disease processes and provide information regarding the possible treatment of diseases. In addition, some prognostic profiles can be formulated by analyzing such antigens.

Summary and Explanation

The /VIEW DAB Detection Kit utilizes biotinylated secondary antibodies to locate the bound primary antibody, followed by the binding of Streptavidin–HRP (horseradish peroxidase) conjugate. The complex is then visualized with hydrogen peroxide substrate and 3, 3' – diaminobenzidine tetrahydrochloride (DAB) chromogen, which produces a dark brown precipitate which is readily detected by light microscopy.

Principles and Procedures

The /VIEW DAB Detection Kit detects specific mouse IgG, IgM and rabbit IgG antibodies bound to an antigen in paraffin embedded or frozen tissues sections. The specific antibody is located by a biotin conjugated secondary antibody. This step is followed by the addition of a streptavidin enzyme conjugate which binds to the biotin present on the secondary antibody. The complex is then visualized utilizing a precipitating enzyme product.

Each step is incubated for a precise time and temperature. At the end of each incubation step, the Ventana automated slide stainer washes the sections to remove unbound material and applies a liquid coverslip which minimizes the evaporation of the aqueous reagents from the slide. Results are interpreted using a light microscope and aid in the differential diagnosis of pathophysiological processes, which may or may not be associated with a particular antigen.